Research Article

Extent, impacts and drivers of oystershell scale invasions in aspen ecosystems

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Abstract

Invasive herbivores that kill foundation tree species pose a major threat to forest ecosystem sustainability. One important foundation tree species in the interior western United States is quaking aspen (*Populus tremuloides* Michx.), which is threatened by recent outbreaks of an invasive insect, oystershell scale (*Lepidosaphes ulmi* Linn.; OSS). OSS outbreaks were first reported in 2016, when OSS began causing dieback and mortality of aspen in wildland forest settings in northern Arizona. Since then, OSS has been observed in other locations across Arizona and in other western states, and recent studies in Arizona have highlighted the threat that OSS poses to aspen sustainability, warranting a comprehensive survey of OSS invasions and their impacts on aspen ecosystems. We sampled aspen populations across Arizona and addressed three questions: (1) What is the geographic extent of OSS in Arizona? (2) What impacts does OSS have on aspen? (3) Which biotic and abiotic factors influence the proportion of aspen stems infested by OSS? OSS was present in 29% of our 220 study plots and had a negative impact on aspen forest health. OSS was associated with crown damage and tree mortality, especially of intermediate-sized, recruiting stems. Climate was the most important driver of OSS infestation, with warmer, drier conditions resulting in significantly more OSS. OSS was also associated with less recent fire, presence of ungulate management strategies (e.g. fenced exclosures) and stands with a greater density of aspen saplings. We conclude by providing OSS monitoring and management recommendations, based on our findings, and emphasise that active management – such as prescribed fire, reduced reliance on ungulate exclosures or thinning – is required to suppress OSS populations and mitigate damage to aspen ecosystems.

Key words: Arizona, armoured scale insect, climate change, invasion ecology, *Lepidosaphes ulmi*, mortality, *Populus tremuloides*, sleeper species

Introduction

Invasive species pose a major threat to sustainability of forest ecosystems (Chornesky et al. 2005). Of particularly high concern are invasive species that kill foundation tree species, resulting in reductions to biodiversity and other ecosystem services (Ellison et al. 2005; Ellison 2019). Unlike keystone species, which are low in abundance but have an outsized influence on ecosystem dynamics, foundation species are common at the landscape scale and often abundant at smaller spatial

scales (Ellison et al. 2005; Ellison 2019). An important foundation tree species in the interior western United States is quaking aspen (*Populus tremuloides* Michx.), which has the widest distribution of any tree species in North America (Little 1971; Perala 1990). Despite its continental abundance, aspen is relatively uncommon along the south-western edge of its range, where aspen occupies less than 2% of forested land (Johnson 1994; Gitlin et al. 2006; Halbritter and Bender 2011; Zegler et al. 2012; Crouch et al. 2023). Additionally, although they occupy a small portion of the landscape, aspen ecosystems have high conservation value because they make disproportionately large contributions to biodiversity and other ecosystem services compared to more abundant conifer-dominated stands (DeByle 1985; Chong et al. 2001; LaMalfa and Ryle 2008; Kuhn et al. 2011; Rogers et al. 2020). Concerningly, aspen in the Interior West is threatened by recent outbreaks of an invasive insect, oystershell scale (*Lepidosaphes ulmi* Linn.; hereafter OSS) (Crouch et al. 2021) (Fig. 1).

OSS is an armoured scale (Hemiptera, Diaspididae) that inserts its stylet through the bark of woody host plants to feed on the fluid of non-vascular cells (Griswold 1925; Beardsley and Gonzalez 1975). This feeding damages host cells, resulting in cell death and cracking of the host's bark under heavy infestations (Beardsley and Gonzalez 1975). Mortality of heavily infested branches, stems and stands may occur (Griswold 1925; Beardsley and Gonzalez 1975). OSS is polyphagous, with around 100 host genera globally (Miller and Davidson 2005b).
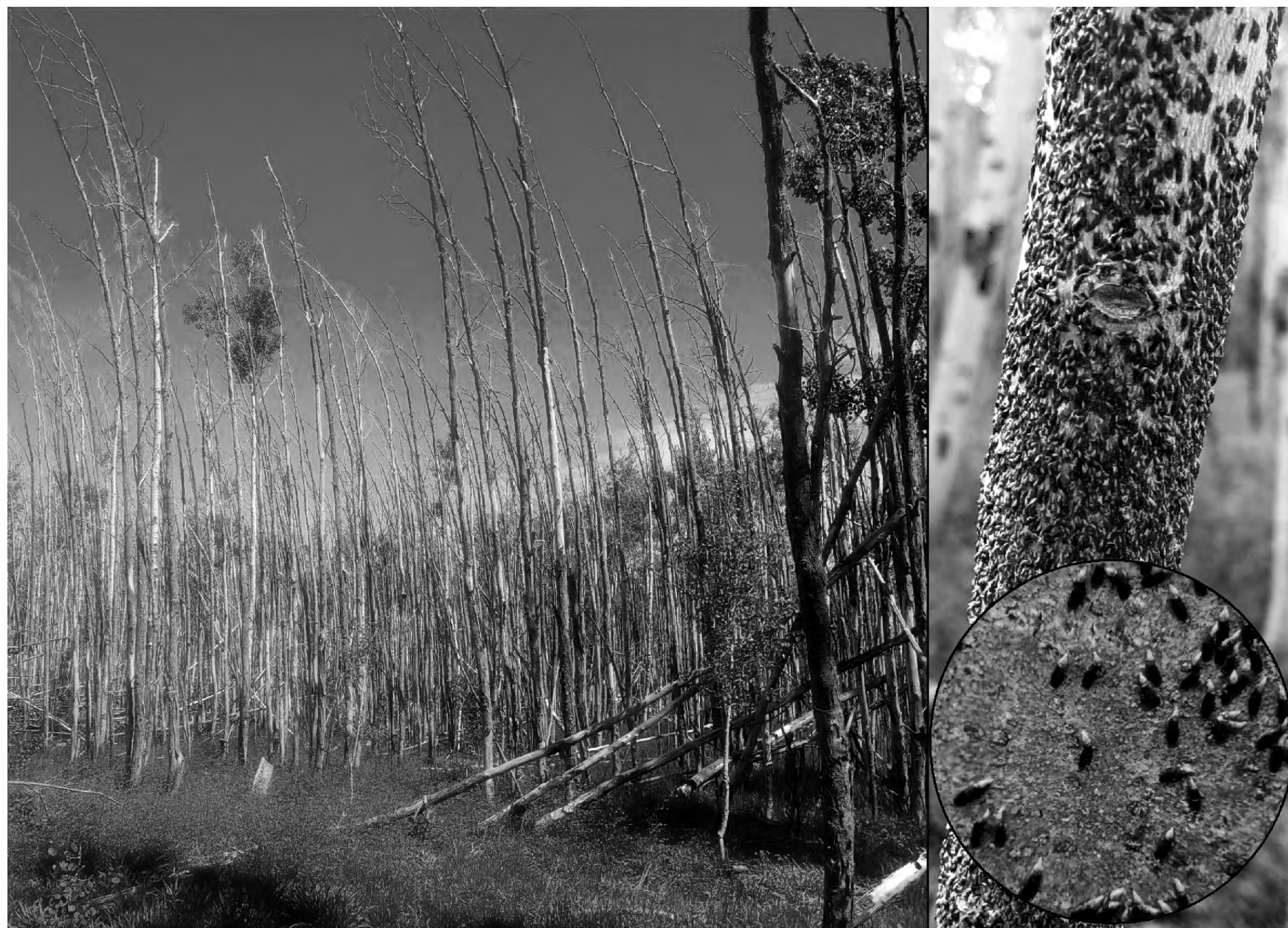

Figure 1. Photo of young aspen stand inside an exclosure (2 m tall fence built to exclude ungulates from browsing aspen) experiencing high levels of dieback and mortality from oystershell scale (OSS). Photos on the right show close-ups of OSS on aspen.

Twelve host genera (*Acer, Alnus, Ceanothus, Cornus, Frangula, Fraxinus, Juglans, Lupinus, Populus, Ribes, Salix* and *Symphoricarpos*) have been documented in the Interior West (Crouch et al. 2021). OSS is most common on woody, deciduous plants, especially those with relatively thin bark (Miller and Davidson 2005b). As an obligate parasite, OSS completes its entire life cycle on woody tissue of living hosts (Samarasinghe 1965; Crouch et al. 2024a). All OSS life stages are sessile, except for first instars (i.e. crawlers), which actively disperse along the host's stem or branches in search of a suitable feeding site (Miller and Davidson 2005b). Once a feeding site is found, OSS will remain there through adulthood (Griswold 1925). In sexually reproducing populations, males shed their tests and fly to mate with females; however, female-only populations occur and reproduce via parthenogenesis (Beardsley and Gonzalez 1975; Miller and Davidson 2005b). It is unclear which reproduction method occurs in the Interior West, although one observation suggests reproduction is predominately asexual (Cranshaw 2013). Questions also remain about OSS dispersal mechanisms and distances, but long-distance crawler dispersal is likely driven by human movement of infested plants, whereas short- and intermediate-distance dispersal are likely driven by wind, potentially with animal assistance (Griswold 1925; Beardsley and Gonzalez 1975; Magsig-Castillo et al. 2010). Crawlers do not actively move more than 1 m due to limited energy reserves and susceptibility to adverse climatic conditions (Beardsley and Gonzalez 1975; Magsig-Castillo et al. 2010).

Although the native range and introduction history of OSS are uncertain, the species was likely transported to North America by European settlers on infested plant material (Griswold 1925; Beardsley and Gonzalez 1975). OSS was first reported as a pest of apple trees (*Malus* spp.) in the 1700s and is now present throughout much of North America, especially in urban and ornamental settings (Griswold 1925; Miller and Davidson 2005b). Despite the species' polyphagous nature, its pervasiveness in North America and its long invasion history, OSS has rarely been a major pest in wildland forest settings (but see Sterrett [1915]; DeGroot [1967]; Houston [2001]). OSS was first reported on aspen in wildland forest settings in Arizona, USA by Fairweather (1992) and Zegler et al. (2012). In both cases, OSS abundance was low and impacts were minimal. However, in 2016, OSS was observed causing dieback and mortality of aspen in wildland forest settings (Grady 2017), and severe outbreaks have since been observed in both areas where Fairweather (1992) and Zegler et al. (2012) first observed OSS. Based on this invasion history, Crouch et al. (2021) suggested that OSS may be a sleeper species, which is defined as a non-native species that establishes successfully, but experiences slow population growth before suddenly awakening when conditions become favourable and experiencing rapid population growth (Groves 1999; Bradley et al. 2018; Frank and Just 2020). It is unclear what led to the awakening of OSS, but recent climatic changes, namely warmer and drier conditions (Williams et al. 2022), are a likely explanation (Crouch et al. 2021). A warming climate may have improved conditions for OSS by increasing the species' fitness and abundance and/or by increasing susceptibility of aspen to infestation and mortality (Frank 2020; Crouch et al. 2021). OSS's role as a sleeper species and its potential interactions with climate change are concerning because continued climate warming may trigger OSS invasions in other areas of aspen's range (Crouch et al. 2021).

Critical to managing any invasive species is baseline information on its occurrence and impacts. The first peer-reviewed report of OSS outbreaks in aspen

ecosystems indicated that OSS is already widespread in northern Arizona (Crouch et al. 2021). Crouch et al. (2021) also observed that OSS only occurred in the lower half of aspen's elevation range (< 2,533 m) and that OSS seemed to be particularly pervasive on and damaging to smaller recruiting stems (stems > 1.37 m tall and < 12.7 cm diameter at breast height [dbh; height = 1.37 m]). Specifically, mortality of recruiting stems was 5–10 times greater than that of overstorey trees in two sites with severe OSS infestations (Crouch et al. 2021). In a survey of aspen populations across Arizona, Crouch et al. (2024b) corroborated those early results by finding that there was significantly less OSS at higher elevations and that OSS was amongst the most important factors limiting aspen recruitment. Both studies also found that OSS was more common in fenced ungulate exclosures (2 m tall fences built to exclude ungulates from browsing on aspen) than outside of them. As recruitment is critical for self-replacement, these initial findings highlight the threat that OSS poses to aspen resilience and sustainability (DeRose and Long 2014; Crouch et al. 2023), warranting a comprehensive survey of OSS and its impacts on aspen in Arizona. We do not know which factors influence OSS's presence across the landscape and what specific impacts OSS has on aspen ecosystems. Obtaining this information is critical for informing management of OSS and mitigating damage caused by this high-impact invasive species. To fill these knowledge gaps, we sampled aspen populations across Arizona and addressed three questions: (1) What is the geographic extent of OSS in Arizona? (2) What impacts does OSS have on aspen? (3) Which biotic and abiotic factors influence the proportion of aspen stems infested by OSS?

Methods

Study area

Our study area encompassed aspen ecosystems across Arizona. Although OSS affects numerous hosts in Arizona (Crouch et al. 2021), our study focused on aspen because the first OSS outbreaks were observed in aspen ecosystems and because of aspen's importance as a foundation species. Aspen ecosystems in Arizona tend to be small (0.1–25 ha in size), especially compared to more northerly latitudes of aspen's range (Zegler et al. 2012). Aspen is limited to relatively high elevations (2,000–3,000 m), where lower temperatures and higher precipitation allow the drought-intolerant species to survive (Perala 1990; Rehfeldt et al. 2009). At lower elevations, small stands of aspen occur on north-facing slopes or in drainages with increased water availability, and as elevation increases, the aspen component tends to be more abundant and less aspect-limited (Rasmussen 1941; Zegler et al. 2012). In addition to occurring alone in small single-species stands, aspen commonly co-occurs with conifers, including ponderosa pine (*Pinus ponderosa* Lawson & C. Lawson var. *scopulorum* Engelm.) and Douglas-fir (*Pseudotsuga menziesii* [Mirb.] Franco var. *glauca* [Beissn.] Franco) at lower elevations, white pine (*Pinus strobiformis* Engelm. or *Pinus flexilis* James var. *reflexa* Engelm.) and white fir (*Abies concolor* [Gord. & Glend.] ex Hildebr.) at mid-elevations and corkbark fir (*Abies lasiocarpa* [Hook.] Nutt. var. *arizonica* [Merriam] Lemmon) and Engelmann spruce (*Picea engelmannii* Parry ex Engelm.) at the highest elevations.

Site selection

We sampled 220 aspen plots that represent the range of conditions under which aspen exists in Arizona (Fig. 2a). These plots were located across seven major areas: North Kaibab (n = 19), South Kaibab (n = 26), Flagstaff (n = 113), Mogollon Rim (n = 13), White Mountains (n = 25), Prescott (n = 17) and Coronado (n = 7) (Fig. 2a). All data were collected during the 2020, 2021 and 2022 growing seasons (June – October), when aspen trees had leaves.

To ensure that we obtained a representative sample of aspen sites and conditions, we stratified sites across four variables – elevation (≤ 2,400 m, > 2,400 m); aspect (north/east, south/west); ungulate management (none, fenced exclosure or jack-straw treatment [large piles of woody debris protecting aspen regeneration from ungulate browse]); and fire history (0–2 years post-fire, 2–20 years post-fire, > 20 years post-fire) – resulting in 24 strata. We first sought to obtain one plot for each stratum, which we accomplished for 21 of the 24 strata, before building out a sample that was proportional to how much aspen occurs in each stratum. We assessed aspen's actual occurrence in each stratum using an observed GIS layer of aspen's range on three national forest ranger districts surrounding Flagstaff (Flagstaff and Mogollon Rim Ranger Districts of the Coconino National Forest; Williams Ranger
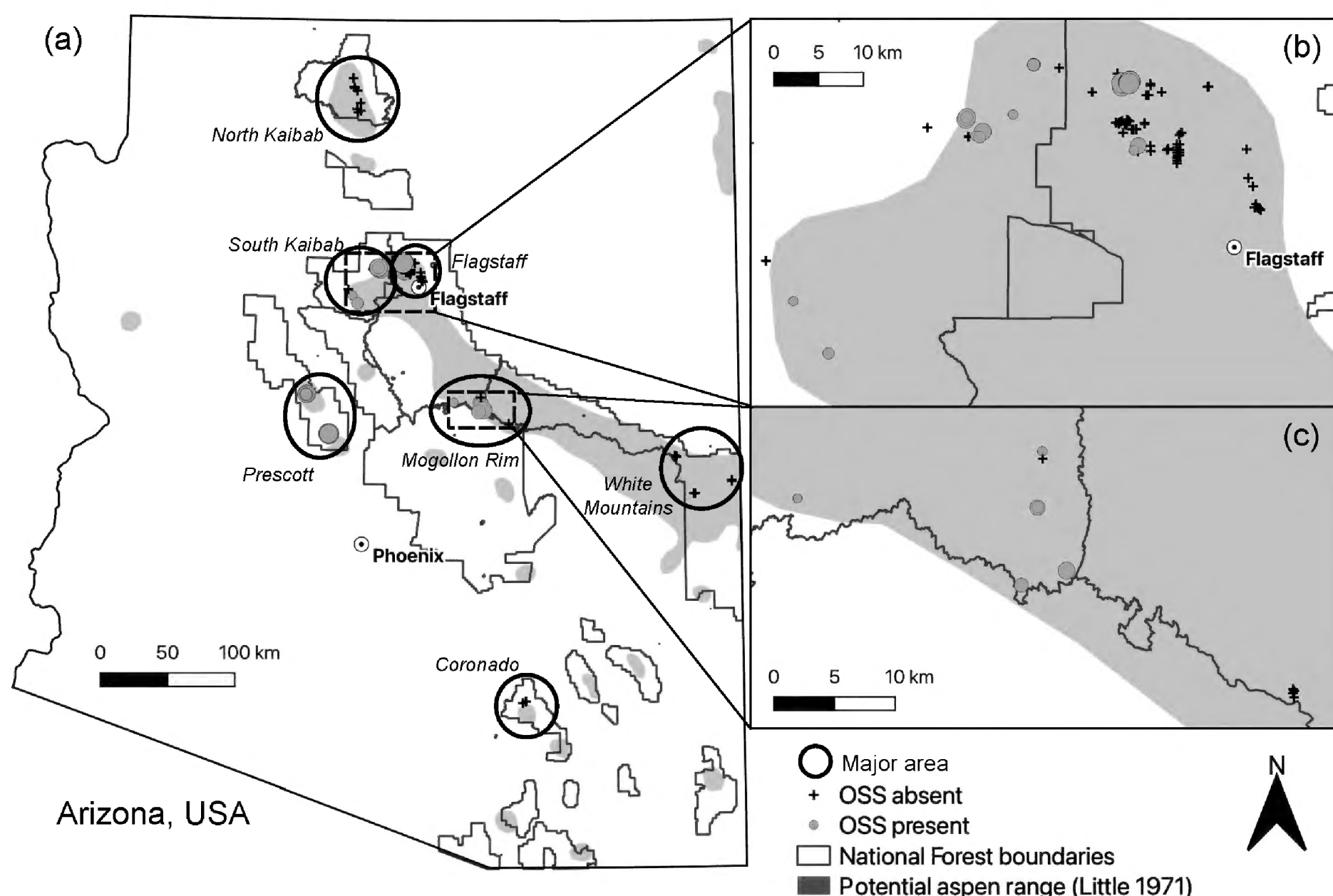

Figure 2. Maps showing OSS presence and absence in **a** 220 study plots across seven major areas (in italics) where aspen occurs in Arizona, USA **b** study plots in the South Kaibab (left of green National Forest boundary line) and Flagstaff major areas (right of green line) and **c** study plots along the Mogollon Rim. These three areas are the only ones where OSS presence varied. OSS was present in all plots around Prescott and was absent in all plots in the North Kaibab, White Mountains and Coronado. Size of OSS presence circles is scaled, based on the proportion of aspen stems infested, with larger circles representing higher infestation rates.

District of the Kaibab National Forest) (DePinte 2018). Although this layer covers only three of the nine ranger districts we sampled, it is the most accurate estimation of where aspen occurs in Arizona because it is a fine-scale layer of aspen's recent presence, based on direct observations from an aircraft (DePinte 2018). We compared the proportion of aspen observed on the landscape, based on area from the GIS layer, to the proportion of aspen plots we sampled, based on the number of plots that fell into each stratum. We succeeded in obtaining a representative sample across elevation, aspect and fire history, with proportions of aspen observed in each stratum versus aspen sampled differing by less than 7% (Crouch et al. 2024b).

When possible, we prioritised re-measurement of existing aspen monitoring plots to reduce the number of redundant plots on the landscape and to facilitate research permission on national forest land. Specifically, we revisited plots previously established by the Coconino National Forest (n = 44), the Apache-Sitgreaves National Forest (n = 5), Zegler et al. (2012) on the Kaibab National Forest (n = 20) and Northern Arizona University's Ecological Restoration Institute on the San Francisco Peaks (n = 12). All four of these networks established plots using stratified or completely random sampling, ensuring the locations of these plots lacked bias. We established the remaining 139 plots by identifying aspen stands that filled target strata, standing on the edge of selected stands, laying out a linear transect longways through those stands and establishing plots every 30 m along the transects. The Coconino National Forest, Apache-Sitgreaves National Forest and Ecological Restoration Institute plots were also established along transects with plot spacings ranging from 100 m to 300 m. The Zegler et al. (2012) plots were established by randomly locating points within known aspen stands and sampling four plots in each cardinal direction 20 m from those points, resulting in plots that were spaced 28.3 m apart.

Field data collection

Each study plot consisted of two fixed-area, circular plots: an overstorey plot (8 m radius) and a nested regeneration plot (4 m radius) sharing the same plot centre (Zegler et al. 2012). We collected GPS coordinates at the centre of each study plot, recorded whether the plot fell in an area of ungulate management (i.e. fenced exclosure or jackstraw treatment) and noted whether there was evidence of recent conifer removal, as indicated by cut conifer stumps present in or directly adjacent to the plot. For a plot to be included in our study, it had to contain at least five live aspen stems between the 8 m overstorey and 4 m regeneration plots combined. In the 8 m overstorey plot, all trees with dbh > 12.7 cm were measured. In the 4 m regeneration plot, all trees > 0.02 cm in height and < 12.7 cm dbh were measured. In the regeneration plot, we classified stems into three size classes adapted from Zegler et al. (2012): short regeneration (< 1.37 m tall), tall regeneration (> 1.37 m tall and < 5.1 cm dbh) and saplings (5.1–12.7 cm dbh). We considered both tall regeneration and saplings to be recruiting stems (Crouch et al. 2024b). For all live aspen, we measured height, dbh (except for regeneration and recruits that were < 1 cm dbh), crown dieback ([percentage of dead branches above the bottom of the tree's live crown] 0%, 1–33%, 34–67%, > 67%), crown ratio (percentage of total tree height occupied by the live crown) and OSS presence and severity. We assessed OSS severity using the system devised by Crouch et al. (2021), which rates OSS severity on each tree from ground level to 6 m. Each tree's stem up to 6 m is divided equally into thirds, and severity is rated for each 2 m section (or shorter for trees

< 6 m tall) on both the north and south sides of the tree. Severity is rated 0–3: 0 = no OSS present, 1 = only a handful of OSS present (trace), 2 = OSS covers < 50% of section (light), 3 = OSS covers > 50% of section (severe). For every dead aspen and live tree species other than aspen, we recorded size class and dbh.

For all live aspen in our study plots, we documented the top three damaging agents present on each tree (Zegler et al. 2012). When more than three damaging agents were present, preference was given to agents with the greatest severity of impact (i.e. most likely to cause dieback and mortality) (Zegler et al. 2012). These damaging agents included insects, diseases, ungulate browse, other animal damage and abiotic damage. For insects and diseases, we grouped individual species into functional groups to facilitate analysis and because some biotic damage agents (e.g. defoliating insects) were impossible to identify based solely on damage signs and symptoms. These groups included sucking and gall-forming insects, bark beetles, wood-boring insects, defoliating insects, canker-causing diseases, foliar and shoot diseases and decay diseases (USDA Forest Service 2013; Steed and Burton 2015). The sucking and gall-forming insects group excluded OSS because we identified and measured OSS separately from other insects. We assessed certain cankers individually because of their potential to have outsized impacts on aspen tree health compared to less pathogenic diseases (Hinds 1985; Zegler et al. 2012; Crouch et al. 2023). The cankers we assessed individually were Cytospora canker (caused by *Cytospora* spp.), Hypoxylon canker (caused by *Entoleuca mammatum* [Wahlenb.] P. Karst), Ceratocystis canker (caused by *Ceratocystis* spp.) and sooty bark canker (caused by *Encoelia pruinosa* [Ellis & Everth.] Torkelson & Eckblad). We lumped all types of abiotic damage together, which included fire scarring of stems, foliar drought scorch and foliar chlorosis. We assessed animal damage to aspen stems, including browse, ungulate barking (i.e. elk chewing aspen bark) and other animal damage. We also indirectly quantified ungulate impacts by counting ungulate scat piles within the 8 m overstorey plot. We identified scat piles by species (i.e. elk [*Cervus canadensis* Erxleben], deer [*Odocoileus hemionus* Rafinesque or *O. virginianus couesi* Coues & Yarrow] or cattle [*Bos taurus* Linn.]) and treated piles from the same species as distinct when piles were clearly separated, contained more than three pellets and differed in colour or size (Bunnefeld et al. 2006; Rhodes and St. Clair 2018).

OSS extent

We assessed OSS presence and absence in each of the 220 study plots to determine the geographic extent of OSS in Arizona. We used descriptive statistics summarising the proportion of plots and trees infested by OSS to further explore geographic patterns of OSS invasions. We also assessed tree-level OSS severity across the four aspen size classes (i.e. short regeneration, tall regeneration, saplings and overstorey trees) to determine if there were differences in susceptibility to OSS. We analysed all data in R version 4.2.1 (R Core Team 2022), using the *dplyr* package (Wickham et al. 2022) for data manipulation and the ggplot2 package (Wickham 2016) for figure creation.

OSS impacts

We assessed OSS impacts on aspen at both the tree and stand levels. At the tree level, we built univariate regressions to quantify the influence of OSS presence and severity on aspen dieback and crown ratio, which are metrics that capture

individual stem health (Schomaker et al. 2007). Tree-level OSS severity was calculated by taking the mean percentage of each category in the rating system (i.e. 1% for 1, 25% for 2 and 75% for 3) and calculating the mean for the six ratings recorded for each tree. We used the *nlme* package (Pinheiro et al. 2022) to fit four linear mixed-effects models with crown dieback and crown ratio as responses, OSS presence and severity as fixed effects and the hierarchical, nested structure of plots (i.e. plots [n = 220] within study sites [n = 87] within minor areas [n = 19] within major areas [n = 7]) as random effects. The random effects in these models and all others in our analysis were specified as random intercepts. Study site refers to a transect or group of plots that were spatially contiguous and occurred within the same strata, with distances between plots in the same site ranging from 30 m to 500 m. Minor area refers to a group of transects or plots in a larger, but still confined area (e.g. an individual mountain or fire footprint). To assess OSS impacts at the stand level, we built univariate regressions between aspen mortality and the proportion of aspen stems infested by OSS in each plot (hereafter referred to as OSS infestation rate). We used the *nlme* package (Pinheiro et al. 2022) to fit six individual linear mixed-effects models with the following response variables: dead aspen basal area, dead aspen density and density of dead aspen in each of the four size classes. We fitted OSS infestation rate as the fixed effect and the hierarchical, nested structure of plots (i.e. plots [n = 64], within study sites [n = 23], within minor areas [n = 9], within major areas [n = 4]) as a random effect. Sample sizes differ for these stand-level models compared to the tree-level models because we used only the 64 plots in which OSS occurred for these stand-level models.

Factors influencing OSS infestation rate

To assess drivers of OSS invasions in aspen ecosystems, we collected data representing an array of biotic (Table 1) and abiotic (Table 2) factors that may influence plot-level OSS infestation rate (i.e. the proportion of aspen stems infested by OSS). In total, we considered 99 variables across eight categories of potential influencing factors: stand structure, ungulates, other damaging agents, fire, management, site factors, soils and climate. We included as many potential influencing factors as possible because we had no prior data on which factors drive OSS invasions. Using tree diameter data, we calculated basal area of stems > 5.1 cm dbh for live aspen, all OSS host species and non-host species (Table 1). We calculated stem densities (trees ha^{-1}) for live aspen, all host species and non-host species across each of four stem size classes (i.e. short regeneration, tall regeneration, saplings and overstorey trees). Using the presence/absence data for all damaging agents on each live aspen stem, we calculated the proportion of stems affected by each agent in each plot (Table 1).

Using GPS coordinates collected at each plot's centre, we calculated elevation, aspect and slope using a 30 m^2 digital elevation model (Table 2). We transformed aspect into a continuous variable ranging from 0–2 with 0 representing southwest (225°) and 2 representing northeast (45°) (Beers et al. 1966). We also calculated heat load and potential annual direct radiation, two indices that assess site-level temperature based on slope, aspect and latitude (McCune and Keon 2002). We assessed fire occurrence in each plot for the past 20 years using wildland fire perimeters obtained from the USDA Forest Service Region 3 GIS database (https://www.fs.usda.gov/detail/r3/landmanagement/gis) and prescribed fire perimeters from national forest staff. We assessed fire severity using data obtained from the

Table 1. List of 33 biotic variables considered as potential influencing factors of plot-level oystershell scale (OSS) infestation rate. Plot-level (n = 220) mean, standard error and range are shown.

Influencing factor	Mean	Std error	Range
Stand structure			
Aspen basal area[a]	10.3	12.4	0–55.9
All hosts basal area	10.3	12.4	0–55.9
Non-host basal area	10.2	14.7	0–78.1
Aspen overstorey density[b]	172	254	0–1,194
Aspen sapling density	354	866	0–6,565
Aspen tall regeneration density	2,399	8,169	0–89,127
Aspen short regeneration density	8,694	17,345	0–136,873
All hosts overstorey density	172	254	0–1,194
All hosts sapling density	356	869	0–6,565
All hosts tall regeneration density	2,460	8,181	0–89,127
All hosts short regeneration density	8,745	17,332	0–136,873
Non-host overstorey density	115	168	0–945
Non-host sapling density	65	180	0–1,592
Non-host tall regeneration density	192	764	0–9,350
Non-host short regeneration density	1,394	3,992	0–33,224
Ungulates			
Browse[c]	0.30	0.31	0–1
Ungulate barking[c]	0.03	0.10	0–0.85
Total ungulate scat[d]	2.6	5.0	0–35
Elk (*Cervus canadensis*) scat	1.3	3.1	0–23
Deer (*Odocoileus hemionus* & *O. virginianus couesi*) scat	1.1	3.5	0–29
Cattle (*Bos taurus*) scat	0.3	1.7	0–20
Damaging agents[c]			
Sucking & gall-forming insects (excluding OSS)	0.09	0.14	0–0.80
Bark beetles	0.01	0.03	0–0.20
Wood-boring insects	0.22	0.21	0–0.83
Defoliating insects	0.60	0.27	0–1
Cytospora canker (caused by *Cytospora* spp.)	0.02	0.05	0–0.34
Hypoxylon canker (caused by *Entoleuca mammatum*)	0.002	0.011	0–0.10
Ceratocystis canker (caused by *Ceratocystis* spp.)	0.02	0.05	0–0.42
Sooty bark canker (caused by *Encoelia pruinosa*)	0.001	0.005	0–0.05
All cankers	0.33	0.26	0–1
Foliar & shoot diseases	0.19	0.24	0–0.94
Decay diseases	0.04	0.08	0–0.67
Other animal damage (excluding browse & barking)	0.01	0.02	0–0.15

[a] basal area = m^2 ha^{-1}
[b] density = trees ha^{-1}
[c] proportion of aspen stems affected by damaging agent
[d] scat = pellet piles/plot

Monitoring Trends in Burn Severity programme (https://www.mtbs.gov/), which provides fire severity data at 30 m resolution. We created categorical variables to represent both fire occurrence and severity in addition to a binary variable for plots that burned twice in the past 20 years (Table 2). Finally, we used GPS coordinates and maps obtained from national forest staff to verify whether plots fell inside areas of ungulate management and conifer removal treatments, and we created binary variables for both ungulate management and conifer removal (Table 2).

Table 2. List of 66 abiotic variables considered as potential influencing factors of plot-level oystershell scale (OSS) infestation rate. Plot-level (n = 220) mean, standard error and range are shown for continuous variables, whereas percentage of plots in each category is shown for categorical variables.

Influencing factor	Mean	Std error	Range
Damaging agents			
Abiotic damage[a]	0.01	0.05	0–0.61
Fire			
Fire strata[b]	1 (14.1%), 2 (22.7%), 3 (63.2%)		
Fire severity[c]	1 (65.5%), 2 (9.1%), 3 (11.4%), 4 (8.2%), 5 (5.9%)		
Burned twice[d]	0 (95.0%), 1 (5.0%)		
Management			
Ungulate management[e]	0 (67.7%), 1 (32.3%)		
Conifer removal[f]	0 (87.7%), 1 (12.3%)		
Site factors			
Elevation (m above sea level)	2,543	237	1,976–3,038
Aspect[g]	0.98	0.73	0–2
Slope (°)	7.9	7.1	0.1–29.7
Heat load (MJ/cm²/yr)	0.98	0.07	0.71–1.08
Radiation (MJ/cm²/yr)	0.96	0.08	0.64–1.09
Major area[h]	1 (3.6%), 2 (51.8%), 3 (6.4%), 4 (9.1%), 5 (8.2%), 6 (11.8%), 7 (11.4%)		
UTM easting	453804	77968	358542–674303
UTM northing	3880092	89053	3589116–4052723
Soils			
Soil order[i]	1 (2.7%), 2 (14.1%), 3 (13.2%), 4 (70.0%)		
Soil pH in H_2O (pH×10)	63.4	2.6	55.4–71.4
Cation exchange capacity (CEC) (mmol(c)/kg at pH 7)	232.9	22.6	176.3–272.15
Nitrogen (cg/kg)	110.0	22.5	80.0–188.3
Soil organic carbon content (dg/kg)	135.2	22.8	93.8–193.9
Bulk density (cg/cm³)	147.5	5.8	130.1–157.8
Sand content (g/kg)	321.7	85.6	187.5–592
Clay content (g/kg)	269.1	51.2	129.7–397.7
Volumetric fraction of coarse fragments (cm³/dm³)	179.2	61.2	75.2–293.0
Climate			
Degree-days below 0 °C	323.9	103.1	109.0–596.0
Degree-days above 5 °C	1,883	377.7	1,215–2,819
Degree-days below 18 °C	3,823	489.9	2,656–4,842
Degree-days above 18 °C	137.1	94.4	24.5–389.0
Degree-days above 10 °C and below 40 °C	909.0	254.3	464.5–1,521.0
Number of frost-free days	181.2	33.8	140.5–265.5
Frost-free period	113.4	29.5	78.0–185.0
Winter temperature (maximum)[j]	6.0	1.6	2.5–10.6
Spring temperature (maximum)	13.8	1.4	10.7–17.8
Summer temperature (maximum)	25.3	1.8	21.2–29.1
Autumn temperature (maximum)	16.6	1.4	13.3–20.3
Winter temperature (minimum)	-7.6	2.2	-10.8– -2.2
Spring temperature (minimum)	-1.2	2.0	-4.4–3.7

Influencing factor	Mean	Std error	Range
Summer temperature (minimum)	9.1	2.1	6.5–14.0
Autumn temperature (minimum)	0.8	2.3	-1.7–6.2
Winter temperature (mean)	-0.8	1.8	-4.2-3.9
Spring temperature (mean)	6.3	1.6	3.2–10.4
Summer temperature (mean)	17.2	1.7	13.9–21.1
Autumn temperature (mean)	8.7	1.6	5.8–12.4
Precipitation as snow (annual)[k]	135.8	56.2	29.5–332.0
Winter precipitation[k]	211.3	75.0	83.5–516.5
Spring precipitation	148.8	34.8	66.5–240.0
Summer precipitation	147.8	69.9	64.5–292.5
Autumn precipitation	130.5	71.8	62.5–366.0
Winter relative humidity[l]	51.0	5.1	44.5–70.5
Spring relative humidity	51.7	3.8	47.0–66.0
Summer relative humidity	53.0	4.8	47.0–63.0
Autumn relative humidity	50.8	5.9	43.0–66.0
Winter Hargreaves reference evaporation[k]	30.0	39.6	0–125.5
Spring Hargreaves reference evaporation	272.1	25.0	192.5–323.0
Summer Hargreaves reference evaporation	463.1	31.2	396.5–535.0
Autumn Hargreaves reference evaporation	224.1	14.1	193.5–263.0
Winter climatic moisture deficit (CMD)[k]	6.2	8.2	0–26.0
Spring climatic moisture deficit (CMD)	163.6	25.0	117.0–216.0
Summer climatic moisture deficit (CMD)	325.6	76.3	177.0–461.5
Autumn climatic moisture deficit (CMD)	139.2	38.6	57.5–199.0
Winter climate moisture index (CMI)[k]	18.5	7.0	5.6–42.7
Spring climate moisture index (CMI)	-3.2	5.2	-14.7–9.6
Summer climate moisture index (CMI)	-29.9	10.3	-48.3– -7.3
Autumn climate moisture index (CMI)	-11.0	8.0	-21.8–15.2
Annual dryness index[m]	0.07	0.02	0.04–0.12
Annual heat moisture index[n]	31.8	8.2	18.1–49.8
Summer heat moisture index[o]	118.8	52.1	45.1–242.4

[a] proportion of aspen stems affected by damaging agent
[b] categorical: 1 (0–2 yrs since fire), 2 (2–20 yrs since fire), 3 (> 20 yrs since fire)
[c] categorical: 1 (unburned in past 20 yrs), 2 (unburned/low), 3 (low), 4 (moderate), 5 (high)
[d] categorical: 0 (burned < 2 times in past 20 yrs), 1 (burned twice in past 20 yrs)
[e] categorical: 0 (no ungulate management), 1 (exclosure or jackstraw)
[f] categorical: 0 (no treatment), 1 (conifer removal)
[g] 0–2 (0 = 225°, 1 = 135° or 315°, 2 = 45°)
[h] categorical: 1 (Coronado), 2 (Flagstaff), 3 (Mogollon Rim), 4 (North Kaibab), 5 (Prescott), 6 (South Kaibab), 7 (White Mountains)
[i] categorical: 1 (Inceptisols), 2 (Mollisols – Borolls), 3 (Mollisols – Ustolls), 4 (Alfisols)
[j] temperature = °C
[k] precipitation, evaporation, CMD, CMI = mm
[l] relative humidity = %
[m] annual dryness index = annual degree days above 5°C ÷ annual precipitation
[n] annual heat moisture index = (annual temperature + 10) ÷ (annual precipitation ÷ 1,000)
[o] summer heat moisture index = warmest month temperature ÷ (summer precipitation ÷ 1,000)
Seasons for climate variables are winter (December – February), spring (March – May), summer (June – August), autumn (September – November).

We obtained soils data from SoilGrids (https://www.isric.org/explore/soilgrids), which provides global soil mapping data at 250 m resolution (Poggio et al. 2021). We used 9 of 12 available soil metrics to capture variables that represent soil moisture (e.g. sand content and bulk density), fertility (e.g. cation exchange capacity, nitrogen and soil organic content), rooting environment (e.g. bulk density, clay content and coarse fragments) and chemical environment (e.g. soil pH) (Table 2). We aggregated mean values for each variable to a depth of 1 m because most lateral aspen roots occur within the first 1 m of the soil (Jones and DeByle 1985b). We obtained climate data from ClimateNA (https://climatena.ca/), which downscales PRISM data (Daly et al. 2008) at 800 m resolution (Wang et al. 2016). We expected climate to be an important driver of OSS infestation rate, so we included an array of climate variables that could potentially influence OSS and aspen. Specifically, we obtained variables representing annual and, when available, seasonal degree-days, temperature, precipitation, humidity, Hargreaves reference evaporation (hereafter evaporation) and drought for the two years preceding the date each plot was sampled (Table 2). We chose two years because repeated observations of OSS in the same field sites across multiple years indicated that most OSS we observed on trees had accumulated in the preceding two years (Crouch et al. 2024a). Although dead OSS are likely capable of lasting even longer on host trees, more recent climate data captures the most recent trends in OSS population growth.

We used random forests, structural equation modelling (SEM) and stand- and tree-level regressions to determine which biotic and abiotic factors drive OSS invasions. First, we used random forests to determine which of the 99 predictor variables had the strongest influence on plot-level OSS infestation rate. Random forests are a useful tool for assessing variable importance in regression and classification settings amongst an array of potential predictors (Breiman 2001). Specifically, we used the *VSURF* package (Genuer et al. 2015), which used 50 random forest runs, each of which was built using 2,000 trees, to rank predictor variable importance. *VSURF* is robust in noisy, high dimensional settings and in the presence of highly correlated predictors (Genuer et al. 2010). *VSURF* outputs a ranked list of variables, based on importance, which is calculated using out-of-bag mean square error for each tree. We used this ranked list of variables when building SEMs and assessing univariate relationships between influencing factors and OSS infestation rate. We also examined the top 10 climate variables in this list to search for climatic thresholds beyond which OSS does not occur in Arizona.

Once we obtained a list of variable importance from *VSURF*, we used SEM to assess how the most important predictors and their interactions affect OSS infestation rate. SEM is an insightful tool for ecological research because it allows the user to build models based on theoretical understanding of an ecological system, resulting in a network of causal, multivariate relationships with a complete accounting of direct and indirect relationships and the relative strengths of those relationships (Grace 2006; Lefcheck 2016). Our first step in building an SEM was to construct an *a priori* model, based on our theoretical understanding of how biotic and abiotic factors might influence OSS. This *a priori* model (Fig. 3) accounted for all 99 variables that potentially influence OSS infestation rate using the eight categories of influencing factors (i.e. stand structure, ungulates, other damaging agents, fire, management, site factors, soils and climate). We then built a "full" SEM, which included the highest ranked variable, based on random forests from each of the eight categories of influencing factors (Tables 1, 2). We used a combination of backward and
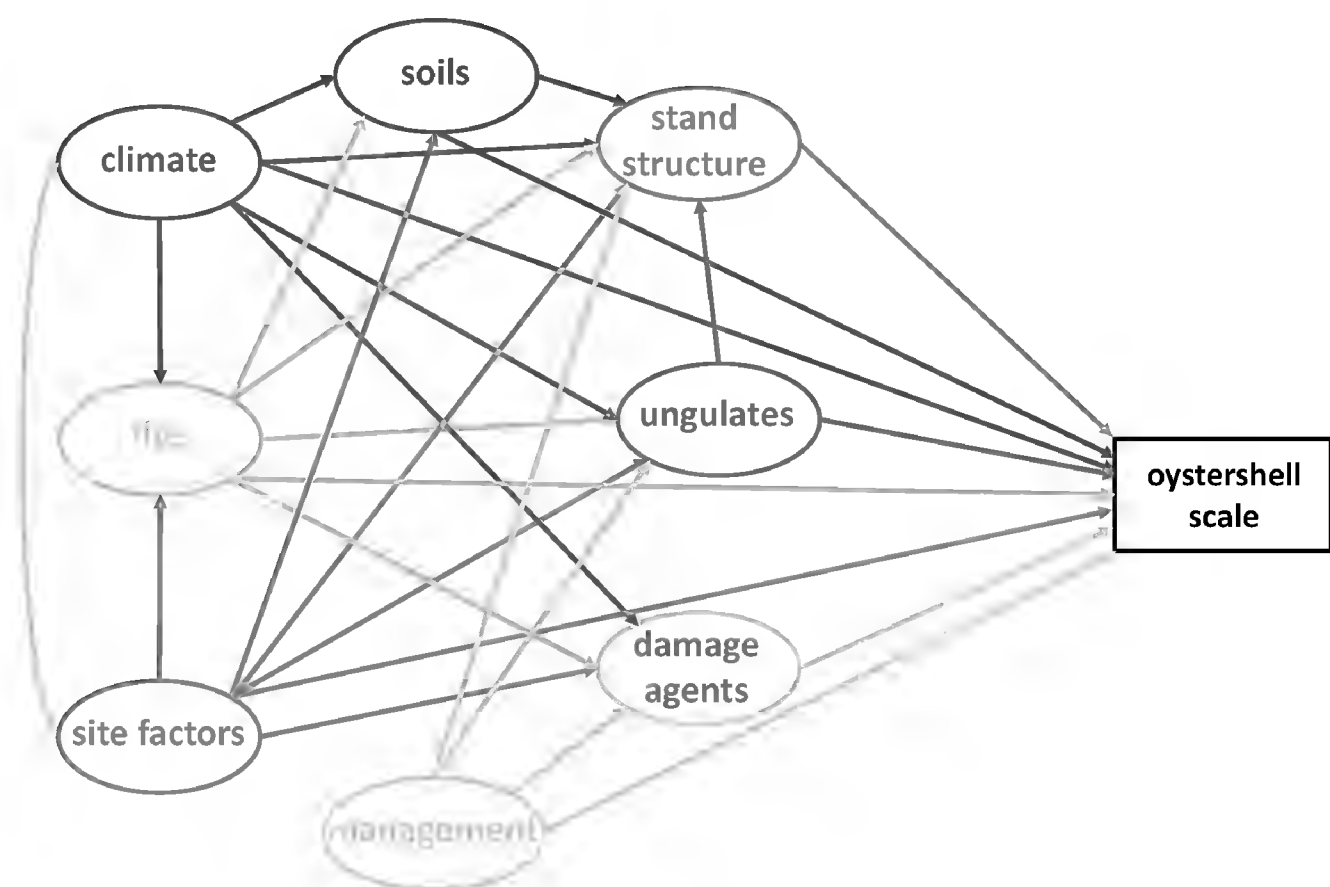
Figure 3. *A priori* structural equation model (SEM) illustrating hypothesised directional relationships amongst influencing factors and plot-level OSS infestation rate. Arrows indicate causal relationships, and colours correspond to each of the eight categories of influencing factors. See Tables 1, 2 for complete lists of measured variables included in each of these eight categories.

forward selection to optimise model fit (using AIC and Fisher's C statistic) and to maximise explanatory power (using R^2 of the response variable). This optimisation process included removing variables with low significance in the model and adding in more than one variable per category (e.g. adding a second climate variable) when two variables from one category had high importance values, based on random forests. We also tested how swapping in one variable to replace another variable of the same category (i.e. replacing fire severity with fire strata) affected the model. To overcome issues with how SEM handles categorical predictors, we set categorical levels as numeric (i.e. 0 and 1 for binary variables or 0–*k* for ordinal variables with *k* levels) and modelled these variables numerically. We used the *piecewiseSEM* package to build SEMs because this package accommodates the use of mixed-effects models (Lefcheck 2016). For the individual regressions that underlie *piecewiseSEM*, we used the *lme4* package (Bates et al. 2015) to fit linear mixed-effects models with the hierarchical, nested structure of plots modelled as random effects.

We also fitted stand- and tree-level regressions to further assess how various factors influence OSS presence and severity. At the stand level, we took the top 25 factors influencing OSS infestation rate based on random forests and built univariate regressions to quantify relationship direction, strength and significance. We used the *nlme* package (Pinheiro et al. 2022) to fit linear mixed-effects models with plot-level OSS infestation rate as the response, the 25 individual influencing factors as fixed effects and the hierarchical, nested structure of plots as random effects. At the tree level, we built univariate regressions to determine the influence of aspen tree size on OSS presence and severity. We used the *nlme* package (Pinheiro et al. 2022) to fit eight linear mixed-effects models with OSS presence and severity as responses, with dbh, height, height-to-diameter ratio and size class as fixed effects and with the hierarchical, nested structure of plots as random effects. As size class is a categorical variable, we used the "anova" function in R (R Core Team 2022) to conduct one-way analysis of variance (ANOVA), allowing us to test for significant differences in OSS

presence and severity amongst the four size classes. When ANOVA found a significant (α = 0.05) difference between size classes, we used the *emmeans* (Lenth 2022), *multcomp* (Hothorn et al. 2008) and *multcompView* (Graves et al. 2019) packages to conduct post-hoc Tukey-adjusted pairwise comparisons and determine which size classes significantly differed. We used these same ANOVA procedures to compare height-to-diameter ratios inside versus outside areas of ungulate management to assess how these treatments influence aspen growth and, in turn, potentially OSS.

Results

Aspen condition

The mean total aspen basal area, including living and standing dead trees, in our 220 study plots was 14.6 m² ha⁻¹ (standard error [SE] = 1.0), of which dead trees made up 29.5% (Table 3). Mean total aspen density was 16,069 trees ha⁻¹ (SE = 1,304), 27.7% of which were dead. Looking at individual stem size classes, study plots contained an average of 239 overstorey aspen ha⁻¹ (SE = 20; 28.2% of which were dead), 519 saplings ha⁻¹ (SE = 79; 31.9% dead), 3,196 tall regeneration stems ha⁻¹ (SE = 626; 24.9% dead) and 12,115 short regeneration stems ha⁻¹ (SE = 1,587; 28.2% dead). Of the 9,965 live aspen stems we sampled, 34.2% of stems had no crown dieback, 44.5% of stems had 1–33% dieback, 13.9% of stems had 34–67% dieback and 7.5% of stems had 67–99% dieback (Table 3). Mean crown ratio of live aspen was 52.0% (SE = 0.5%).

Table 3. Summary data for aspen stand structure, crown condition and OSS presence. Means and standard errors of variables representing live and dead aspen basal area, live and dead aspen density in different stem size classes, live aspen crown ratio and dieback and OSS presence at the plot and tree levels. For categorical variables, percentage of plots or trees in each level are shown.

Variable	Mean	Std error
Live aspen basal area (m² ha⁻¹)	10.3	0.8
Dead aspen basal area (m² ha⁻¹)	4.3	0.4
Total live aspen density (trees ha⁻¹)	11,618.5	1,304.2
Total dead aspen density (trees ha⁻¹)	4,450.5	704.2
Live aspen short regeneration density (trees ha⁻¹)	8,693.8	1,169.4
Dead aspen short regeneration density (trees ha⁻¹)	3,420.9	674.6
Live aspen tall regeneration density (trees ha⁻¹)	2,399.1	550.8
Dead aspen tall regeneration density (trees ha⁻¹)	796.7	126.6
Live aspen sapling density (trees ha⁻¹)	353.6	58.4
Dead aspen sapling density (trees ha⁻¹)	165.5	42.6
Live aspen overstorey density (trees ha⁻¹)	172.0	17.1
Dead aspen overstorey density (trees ha⁻¹)	67.4	8.6
Aspen crown ratio (%)	52.0	0.5
Aspen crown dieback (categorical)		
0% dieback	34.2	0.5
1–33% dieback	44.5	0.5
34–67% dieback	13.9	0.3
68–99% dieback	7.5	0.3
Plot-level OSS presence (categorical)		
OSS absent	70.9	3.1
OSS present	29.1	3.1
Tree-level OSS presence (categorical)		
OSS absent	89.3	0.3
OSS present	10.7	0.3

OSS extent

OSS was present in 29% of study plots and occurred in four of seven major areas where aspen occurs in Arizona: South Kaibab, Flagstaff, Prescott and Mogollon Rim (Fig. 2a). OSS was not found in study plots we sampled in the North Kaibab, White Mountains or Coronado major areas. Prescott had the highest plot-level rate of infestation, with OSS present in all 17 plots. South Kaibab had 65.4% of plots infested (Fig. 2b), Mogollon Rim had 53.4% of plots infested (Fig. 2c) and Flagstaff had 20.4% of plots infested (Fig. 2b). Of the 9,965 live aspen stems we sampled, 10.7% were infested with OSS (Table 3). Tree-level rates of infestation across major areas were consistent with plot-level rates of infestation. When looking at all plots, not just those in which OSS occurred, Prescott had the highest proportion of trees infested (60.3%), followed by South Kaibab (20.8%), Mogollon Rim (16.2%) and Flagstaff (7.6%).

OSS infested aspen stems of all sizes, although there was a higher likelihood of infestation on trees taller than 1.37 m (i.e. tall regeneration stems and larger) (Fig. 4). Using the OSS severity rating, 6.9% of all live aspen stems we sampled had a mean rating of trace (only a handful of OSS present), 2.3% were light (OSS covers < 50% of measured tree surface) and 1.4% were severely infested (OSS covers > 50% of measured tree surface). In the 64 plots where OSS was present, mean severity ratings were 34.5% trace, 11.6% light and 7.2% severe. Patterns of OSS severity across stem size classes generally followed those of OSS presence, with more severe infestations occurring as rate of presence increased (Fig. 4).
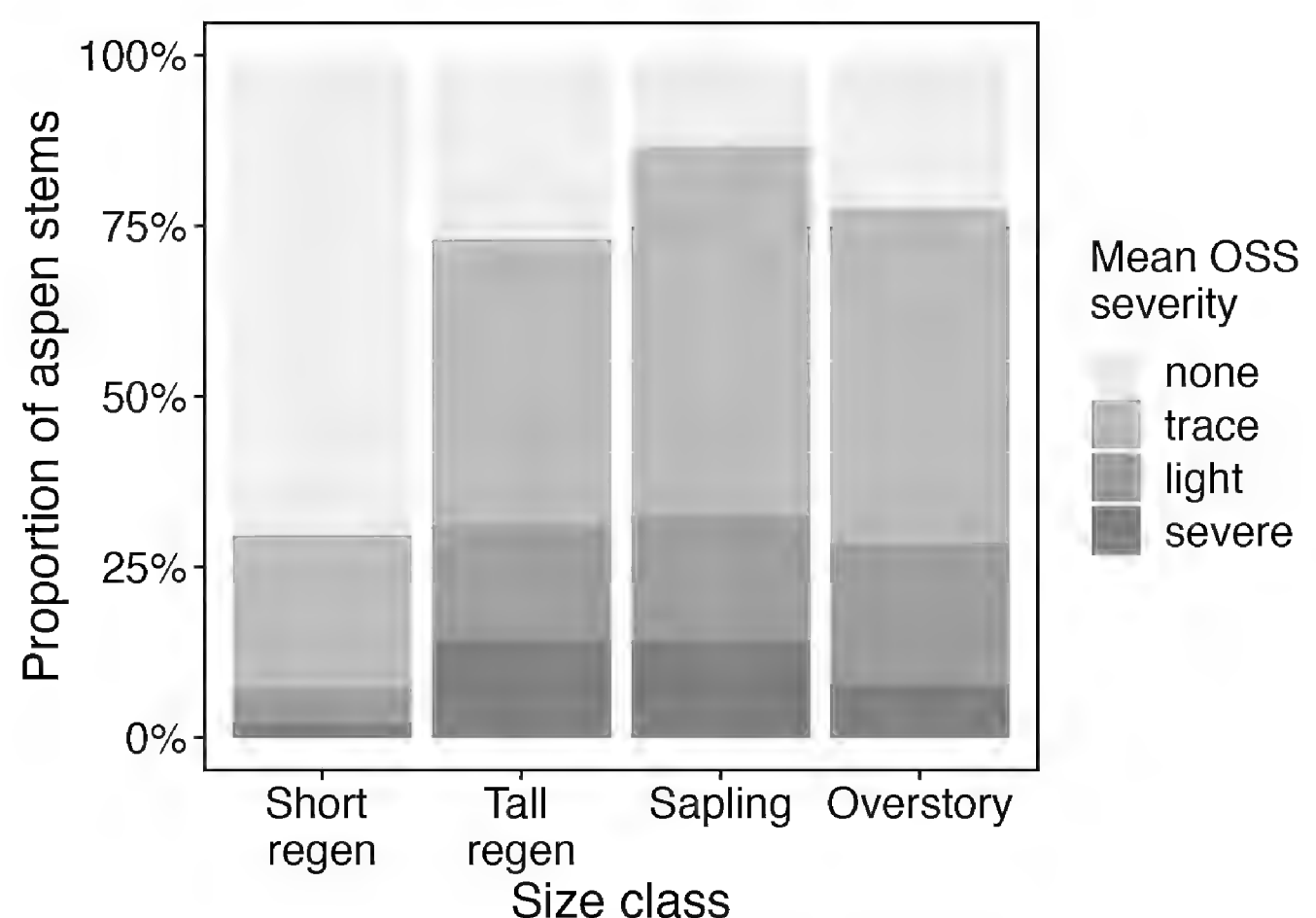

Figure 4. OSS severity across four aspen stem size classes. Data shown were taken only from the 64 study plots in which OSS was observed and include only live trees. OSS severity was assessed using the rating system devised by Crouch et al. (2021), which rates OSS severity on each tree from ground level to 6 m. Each tree's stem up to 6 m is divided into thirds, and severity is rated for each 2 m section (or shorter for trees < 6 m tall) on both the north and south sides of the tree. The ratings are as follows: none (no OSS present), trace (only a handful of OSS present), light (OSS covers < 50% of section), severe (OSS covers > 50% of section).

OSS impacts

OSS presence at the tree level was significantly ($p < 0.001$) associated with reduced aspen crown ratio and increased crown dieback, based on univariate regression (Table 4). Trees infested with OSS were associated with a 9.8% reduction in crown ratio and a 0.5 unit increase in dieback compared to trees without OSS. Dieback was assessed on a categorical scale from 0 to 3 (0 = 0% dieback, 1 = 1–33% dieback, 2 = 34–67% dieback, 3 = 67–99% dieback), so a 0.5 unit increase on the categorical scale equates to a roughly a 16% increase in dieback. Tree-level OSS severity was also significantly ($p < 0.001$) associated with reduced crown ratio and increased dieback (Table 4). Tree-level OSS severity ranged from 0 to 0.75, with 0 indicating a tree without OSS and 0.75 indicating a tree with the highest possible severity rating (OSS covering > 50% of all six stem sections rated). Thus, an increase in OSS severity of 0.1 equates to a 10% increase in OSS infestation of the tree's stem up to 6 m in height. A 0.1 unit increase in OSS severity was associated with a 3.9% reduction in crown ratio and a 0.2 unit increase in dieback, which equates to roughly 6.6% dieback. Despite the high degree of significance for all four of these univariate relationships, OSS presence and severity explained a low proportion of the variance observed in aspen crown ratio and dieback ($R^2 \leq 0.03$).

For the 64 study plots in which OSS was present, plot-level OSS infestation rate was significantly ($p = 0.019$) associated with increased dead aspen basal area (Table 5). An increase in OSS infestation rate of 1, which represents the difference between no stems infested by OSS and all stems infested by OSS, was associated with an increase in dead aspen basal area of 5.7 m² ha⁻¹. For context, mean basal area of living aspen in these 64 plots was 11.4 m² ha⁻¹ (SE = 1.5) and in all 220 study plots was 10.3 m² ha⁻¹ (SE = 12.4). We also assessed the influence of OSS infestation rate on five measures of dead aspen density: total dead aspen and density of each of the four size classes. None of these univariate relationships was significant, although the tall regeneration ($p = 0.054$) and sapling ($p = 0.061$) models approached significance (Table 5). An increase in OSS infestation rate of 1 was associated with 1,654 more dead tall regeneration stems ha⁻¹ and 532 more dead saplings ha⁻¹. For context, there were 1,579 living tall regeneration stems ha⁻¹ (SE = 167) and 867 living saplings ha⁻¹ (SE = 387) on average in the 64 plots where OSS occurred and 2,399 tall regeneration stems ha⁻¹ (SE = 8,169) and 354 living saplings ha⁻¹ (SE = 866) across all study plots. The models for total dead aspen density, density of dead short regeneration and density of dead overstorey trees were insignificant ($p \geq 0.350$).

Table 4. Univariate relationships between two measures of aspen stem health (crown ratio and dieback) and OSS presence and severity at the tree level.

Response	Predictor	Coefficient	Std error	p value	Marginal R²
Aspen crown ratio (%)	OSS presence[a]	-9.83	1.25	< 0.001	0.012
Aspen crown ratio (%)	OSS severity (%)	-38.87	4.33	< 0.001	0.009
Aspen crown dieback[b]	OSS presence[a]	0.52	0.04	< 0.001	0.031
Aspen crown dieback[b]	OSS severity (%)	2.06	0.14	< 0.001	0.024

These relationships are based on linear mixed models. Marginal R² is based solely on the model's fixed effects, which were either OSS presence or severity.
[a] 0–1 (0 = OSS absent, 1 = OSS present)
[b] 0–3 (0 = 0% dieback, 1 = 1–33% dieback, 2 = 34–67% dieback, 3 = 68–99% dieback)

Table 5. Univariate relationships between six measures of dead aspen density and plot-level OSS infestation rate (i.e. proportion of stems infested by OSS). These models were fitted using data only from the 64 study plots in which OSS was present.

Response	Predictor	Coefficient	Std error	p value	Marginal R^2
Dead aspen basal area[a]	OSS (%)	5.74	2.35	0.019	0.108
Total dead aspen[b]	OSS (%)	1,029.67	1,644.03	0.535	0.005
Dead aspen short regeneration[b]	OSS (%)	-646.59	961.49	0.505	0.005
Dead aspen tall regeneration[b]	OSS (%)	1,654.33	834.21	0.054	0.076
Dead aspen saplings[b]	OSS (%)	532.32	275.75	0.061	0.070
Dead overstorey aspen[b]	OSS (%)	33.15	35.04	0.350	0.015

These relationships are based on linear mixed models. Marginal R^2 is based solely on the model's fixed effect, which was plot-level OSS infestation rate.
[a] m^2 ha^{-1}
[b] trees ha^{-1}

Factors influencing OSS infestation rate

We considered 99 potential factors influencing plot-level OSS infestation rate (i.e. proportion of aspen stems infested by OSS), and random forests indicated the five most important influences were autumn evaporation, elevation, degree-days between 10 °C and 40 °C, winter climate moisture index (CMI) and autumn precipitation (Table 6). Based on univariate relationships between the top 25 most important influencing factors and OSS infestation rate, the five strongest influencing factors were maximum winter temperature ($R^2 = 0.43$; $p < 0.001$), winter evaporation ($R^2 = 0.41$; $p < 0.001$), maximum spring temperature ($R^2 = 0.30$; $p < 0.001$), elevation ($R^2 = 0.26$; $p < 0.001$) and minimum spring temperature ($R^2 = 0.21$; $p = 0.004$).

The optimal SEM for plot-level OSS infestation rate (AIC = 861.6; Fisher's C = 1.018 with $p = 0.907$ [high p value indicates better fit]; response marginal $R^2 = 0.53$, conditional $R^2 = 0.88$ [marginal includes only fixed effects, conditional includes both fixed and random effects]) included seven influencing factors: autumn evaporation, winter CMI, maximum winter temperature, elevation, fire strata, live aspen sapling density and presence of ungulate management (Fig. 5). Based on this SEM, all influencing factors, except for winter CMI and elevation, had a significant ($p < 0.05$) direct effect on OSS infestation rate. Autumn evaporation had a negative direct effect (effect size = -0.33; $p = 0.011$) on OSS infestation rate, whereas maximum winter temperature (0.60; $p = 0.011$), fire strata (0.30; $p < 0.001$), live aspen sapling density (0.13; $p = 0.001$) and presence of ungulate management (0.26; $p < 0.001$) had positive direct effects. Fire strata and ungulate management were categorical variables (Table 2), and SEM indicated that less recent fire and presence of ungulate management resulted in a greater OSS infestation rate. All three climate variables had significant ($p \leq 0.003$) influences on fire strata, with recent fire being driven by less autumn precipitation, higher winter CMI and higher maximum temperatures in winter. Thus, more autumn evaporation indirectly led to more OSS, while higher winter CMI and maximum winter temperatures indirectly led to less OSS. More autumn evaporation and higher winter CMI also resulted in significantly ($p \leq 0.003$) fewer aspen saplings, resulting in both climate variables having an additional negative indirect effect on OSS infestation rate. Finally, aspen sapling density was significantly ($p < 0.001$) lower at higher elevation, resulting in a negative indirect effect of elevation on OSS infestation rate.

Table 6. Relationships between OSS infestation rate and its most important influencing factors based on random forests. Top 25 most important variables influencing plot-level OSS infestation rate, based on 50 random forest runs, each of which was built using 2000 trees. Univariate relationships between influencing factors and OSS infestation rate were based on linear mixed models. See Tables 1, 2 for lists of all influencing factors considered.

Random forests		Univariate regressions		
Rank	Influencing factor	Coefficient	Marginal R^2	*p* value
1	autumn evaporation	-0.003	0.014	0.195
2	elevation	< -0.001	0.263	< 0.001
3	degree-days 10–40 °C	< 0.001	0.197	0.003
4	winter CMI	0.005	0.015	0.302
5	autumn precipitation	0.002	0.168	0.002
6	winter evaporation	0.006	0.413	< 0.001
7	winter temp (max)	0.120	0.428	< 0.001
8	wood boring insects	0.121	0.007	0.034
9	winter precipitation	< -0.001	0.002	0.755
10	clay	< 0.001	0.003	0.638
11	degree-days < 0 °C	-0.001	0.190	0.003
12	snow	-0.002	0.146	0.001
13	spring temp (max)	0.105	0.301	< 0.001
14	spring CMD	0.004	0.109	0.002
15	spring evaporation	0.002	0.037	0.085
16	aspen saplings ha^{-1}	< 0.001	0.018	0.001
17	host saplings ha^{-1}	< 0.001	0.018	0.001
18	degree-days > 5 °C	< 0.001	0.198	0.003
19	spring temp (min)	0.071	0.210	0.004
20	host regeneration ha^{-1}	< -0.001	< 0.001	0.455
21	UTM easting	< -0.001	0.088	0.170
22	other animal damage	0.070	< 0.001	0.891
23	summer temp (mean)	0.068	0.174	0.005
24	aspen regeneration ha^{-1}	< -0.001	< 0.001	0.458
25	fire severity	-0.014	0.003	0.463

Marginal R^2 is based solely on the model's fixed effect, which was the influencing factor shown in each row.

To further assess the relationship between climate and OSS, we searched for thresholds using the top 10 climate variables that random forests indicated were the most important influences of plot-level OSS infestation rate. We identified clear elevational and climatic thresholds beyond which OSS does not occur in Arizona (Fig. 6). OSS was not observed in plots that exceeded 2,545 m in elevation, received greater than 152 mm of snow annually, experienced maximum winter temperatures below 5.25 °C, experienced maximum spring temperatures below 13.25 °C and had fewer than 825 degree-days between 10 °C and 40 °C (calculated as accumulated temperature difference from the degree-day threshold, rather than a true accumulation of degree-days [Wang et al. 2016]). In addition, OSS was not observed in plots with less than 218 mm of autumn evaporation, except for one plot which had 202.5 mm of evaporation and a single aspen stem infested with OSS.

We also assessed univariate relationships between tree-level OSS presence and severity and four measures of aspen stem size: size class, height, dbh and
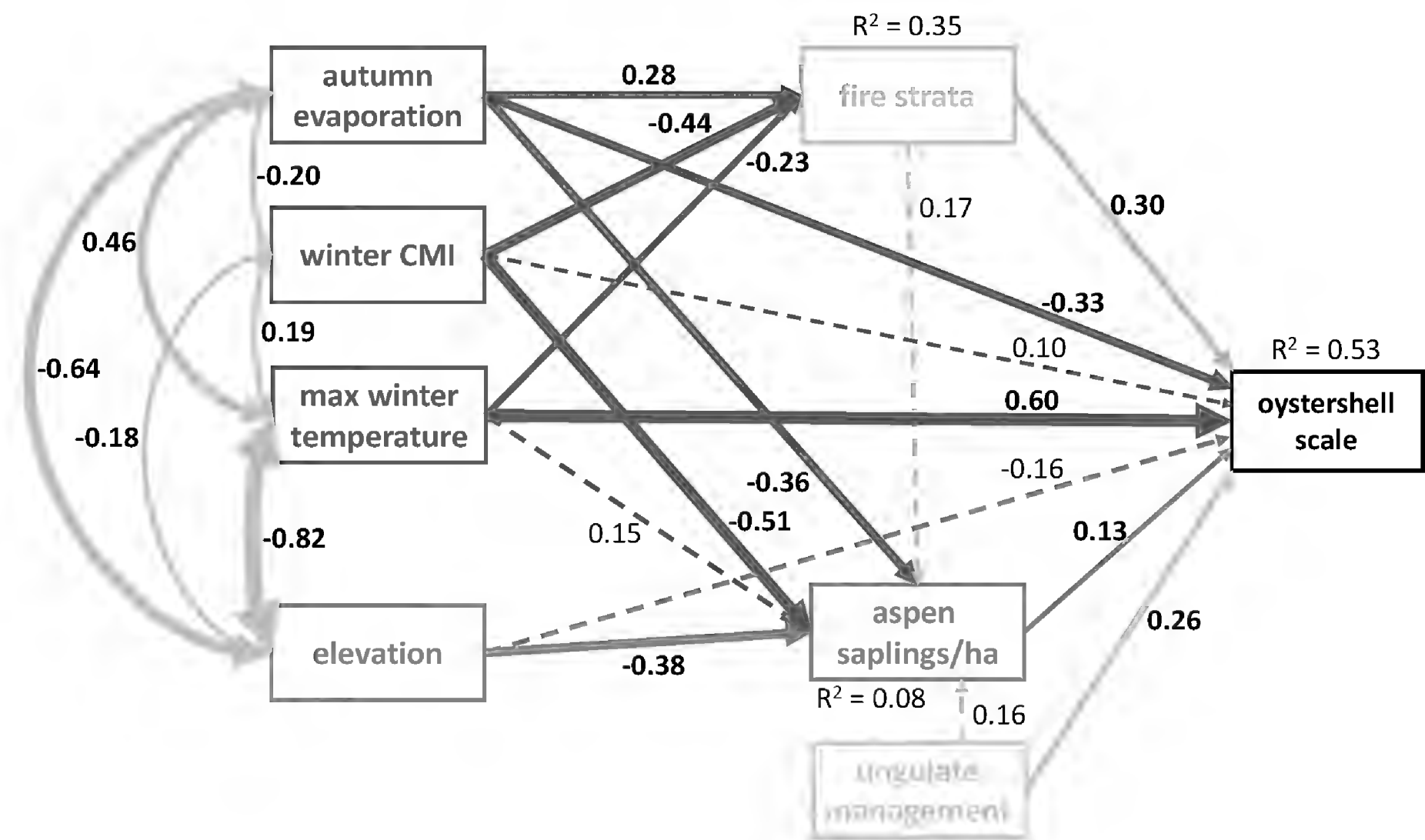
Figure 5. Optimal SEM for OSS infestation rate that minimised AIC and maximised response R^2. Significant ($p < 0.05$) path coefficients are shown in bold, and their corresponding paths are depicted as solid lines. In contrast, insignificant coefficients are not in bold, and their corresponding paths are shown as dashed lines. Path thickness indicates strength of its coefficient, with wider paths indicating stronger relationships.

height-to-diameter ratio. One-way ANOVA indicated that there were significant differences ($p < 0.001$) in OSS presence and severity amongst the four stem size classes (Table 7). OSS presence was significantly greater on overstorey trees and saplings compared to tall and short regeneration, and OSS presence was significantly greater on tall regeneration than short regeneration. Saplings and tall regeneration had significantly greater OSS severity than overstorey trees and short regeneration, while overstorey trees had significantly greater OSS severity than short regeneration. OSS presence and severity significantly ($p < 0.001$) increased with increasing height and decreasing dbh (Table 7). In addition, OSS presence and severity significantly ($p \leq 0.042$) increased with increasing aspen height-to-diameter ratio, also known as slenderness, and one-way ANOVA indicated that height-to-diameter ratios were significantly ($p < 0.001$) greater inside areas of ungulate management than outside these areas. Similar to the tree-level crown ratio and dieback results, aspen stem size explained a low proportion of the variance observed in OSS presence and severity (marginal $R^2 \leq 0.02$) (Table 7).

Discussion

OSS extent

OSS is widely distributed throughout aspen ecosystems in central Arizona (Fig. 2a), confirming the initial report of OSS outbreaks in the region (Crouch et al. 2021). This study added plots in three areas not surveyed by Crouch et al. (2021) – North Kaibab, White Mountains and Coronado – none of which had OSS.
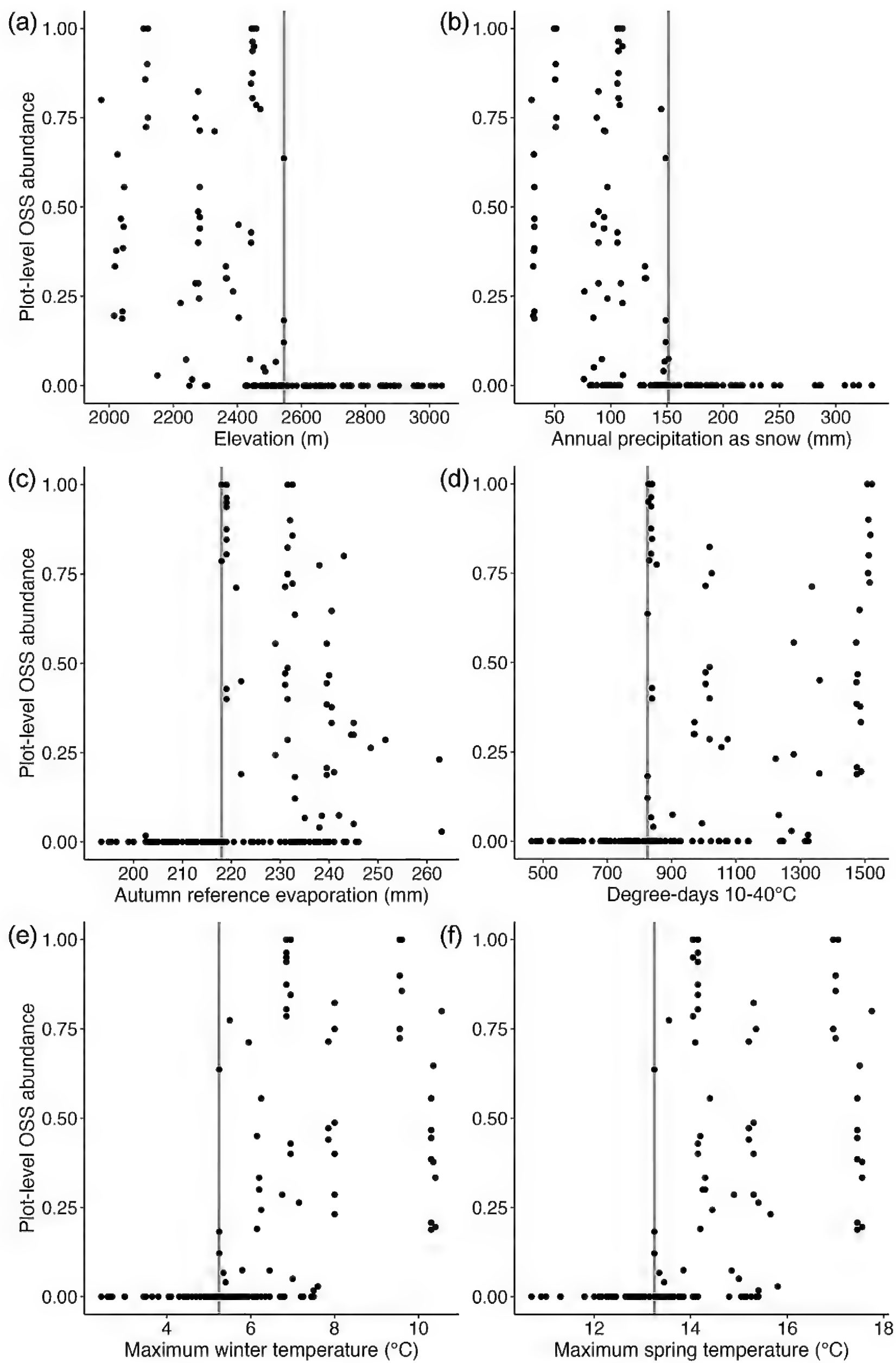
Figure 6. Elevation and climate thresholds beyond which OSS does not occur in aspen ecosystems in Arizona. Relationships between plot-level OSS infestation rate and **a** elevation **b** snowfall **c** autumn evaporation **d** degree-days above 10 °C and below 40 °C **e** maximum winter temperature and **f** maximum spring temperature. Red lines indicate thresholds above or below which OSS does not occur in aspen ecosystems in Arizona.

Table 7. Univariate relationships between tree-level OSS presence and severity and four measures of aspen stem size (size class, height, dbh and height-to-diameter ratio).

Response	Predictor	Coefficient	Std error	*p* value	Marginal R²
OSS presence [a]	size class SR	-12.246 c	1.936	< 0.001	0.014
	TR	1.752 b	0.189	< 0.001	
	S	2.905 a	0.279	< 0.001	
	O	2.904 a	0.280	< 0.001	
OSS severity (%)	size class SR	0.010 c	0.008	0.239	0.021
	TR	0.027 a	0.002	< 0.001	
	S	0.035 a	0.004	< 0.001	
	O	0.011 b	0.003	< 0.001	
OSS presence[a]	height (m)	0.006	0.001	< 0.001	0.004
OSS severity (%)	height (m)	0.001	< 0.001	< 0.001	0.002
OSS presence[a]	dbh (cm)	-0.003	0.001	< 0.001	0.005
OSS severity (%)	dbh (cm)	-0.001	< 0.001	< 0.001	0.012
OSS presence[a]	height:diameter (m)	0.458	0.166	0.006	0.001
OSS severity (%)	height:diameter (m)	0.114	0.056	0.042	0.001

These relationships are based on linear mixed models. Aspen size class abbreviations: SR (short regeneration, < 1.37 m tall), TR (tall regeneration, > 1.37 m tall and < 5.1 cm dbh), S (saplings, 5.1–12.7 cm dbh), O (overstorey trees, > 12.7 cm dbh). Different letters after coefficients indicate significant differences amongst size classes, based on post-hoc Tukey-adjusted pairwise comparisons. Marginal R² is based solely on the model's fixed effects, which was aspen size class, height, dbh or height-to-diameter ratio.
[a] 0–1 (0 = OSS absent, 1 = OSS present)

We also added plots in the South Kaibab, Flagstaff, Prescott and Mogollon Rim major areas, identifying more sites where OSS occurs in these areas than were initially reported (Grady 2017; Crouch et al. 2021). Aspen ecosystems we sampled in the North Kaibab and White Mountains may be free of OSS because wetter, colder climates in those areas (Suppl. material 1) may be unsuitable for OSS, as we discuss later. All plots we sampled in both these areas were above 2,545 m in elevation, which was the threshold above which we did not observe OSS anywhere in Arizona (Fig. 6a). Alternatively, these areas may have evaded OSS invasions for a different, unknown reason. For example, OSS spread at the landscape scale is likely facilitated by its ability to infest a wide array of hosts in addition to aspen (Miller and Davidson 2005b; Crouch et al. 2021). However, our ability to determine how other host species facilitate OSS invasions of aspen ecosystems is limited due to the lack of fine-scale mapping of those species' ranges, particularly understorey hosts, such as *Ceanothus* spp. Unlike the North Kaibab and White Mountains, aspen ecosystems on the Coronado tend to occur on relatively warmer, drier sites (Suppl. material 1), but OSS may have been absent in our Coronado study plots because they were all located in areas burned the previous year by the 2020 Bighorn Fire. Importantly, our sampling was not exhaustive and was restricted to aspen ecosystems, so OSS may occur in the North Kaibab, White Mountains and Coronado. Although some of our study plots contained non-aspen tree species that are OSS hosts (Table 1), few of these were infested, and our monitoring did not account for presence of understorey hosts. More extensive monitoring in the areas where we did not find OSS, including on hosts other than aspen, is warranted.

Prescott had the highest rates of OSS infestation of the seven major areas we studied, with 100% of plots (n = 17) and 60.3% of live aspen in the region being

infested. This is concerning because Prescott also had the highest levels of sustainable aspen recruitment, defined as the number of recruits needed for successful self-replacement of the existing overstorey (Crouch et al. 2024b), so OSS outbreaks in this area might eventually counteract successful recruitment. One reason why there is so much aspen recruitment in Prescott is because elk populations, which significantly inhibit aspen recruitment (Beschta and Ripple 2010; Fairweather et al. 2014; Crouch et al. 2023, 2024b), do not occur in the area. Therefore, ungulate exclosures, which also do not exist in the area, do not facilitate OSS invasions of aspen ecosystems in Prescott as they seem to do in other areas of Arizona. Instead, Prescott may have more OSS than other areas because it has been present in the area for longer. The earliest report of OSS occurring on wildland aspen in Arizona was in Prescott approximately 30 years before our study occurred (Fairweather 1992). Another possible explanation for why OSS is so abundant in Prescott is because aspen sites in this area tend to experience warmer temperatures than aspen in other parts of Arizona (Suppl. material 1). As we discuss below, warmer temperatures are a significant driver favouring OSS.

OSS impacts

OSS negatively affected aspen health at both the tree and stand levels. Aspen trees infested with OSS had significantly lower crown ratios and higher dieback, indicating reduced stem health. As OSS infestations became more severe, crown ratio significantly decreased, and dieback significantly increased (Table 4). Despite the significance of these observed effects, OSS explained a low proportion of variance in aspen crown ratio and dieback ($R^2 \leq 0.03$), indicating that OSS is one of many factors influencing aspen health. Crown damage was likely caused by the feeding behaviour of OSS, which extracts fluid from the host plant's non-vascular cells (Griswold 1925; Beardsley and Gonzalez 1975). Although crown dieback is a common symptom associated with feeding of armoured scales on the bark of woody hosts, the exact mechanism that causes crown damage is not well understood (Miller and Davidson 2005a). One exception is San Jose scale (*Diaspidiotus perniciosus* Comstock), for which feeding on woody tissue has been shown to impair growth of affected cells and alter the number, size and arrangement of xylem and phloem cells (Beardsley and Gonzalez 1975). If similar cell damage is caused by OSS feeding, then crown damage may be the result of impaired water transport and increased vulnerability to embolism (Hillabrand et al. 2019). OSS might also damage aspen by reducing photosynthetic capacity because aspen bark is photosynthetic (Jones and DeByle 1985b). Alternatively, OSS feeding may compromise stem health by causing trees to allocate resources from growth to defence (Cope et al. 2021). At the stand level, plots with more stems infested by OSS had significantly greater dead aspen basal area, with a 10% increase in OSS infestation rate equating to an increase of 0.57 m² ha⁻¹ in dead aspen basal area (Table 5). Although density of total dead aspen stems and density of dead stems in each of the four size classes had insignificant relationships with OSS infestation rate, the models for dead tall regeneration and saplings approached significance, suggesting that these intermediate-sized, recruiting stems may be more susceptible to OSS than regeneration and overstorey trees (Crouch et al. 2024b).

Density of aspen saplings and saplings of all host species were significantly associated with increased plot-level OSS infestation rate (Table 6), providing additional

evidence that intermediate-sized, recruiting stems seem to be most susceptible to OSS. At the tree level, OSS infestations were more severe on tall regeneration and saplings and were associated with taller and thinner stems (Table 7). We hypothesise that short regeneration had such low infestation rates because these stems remain shorter than 1.37 m for only a few years (Jones and Schier 1985), which reduces the likelihood of OSS finding them before they grow into taller size classes. Short regeneration may also be more likely to evade OSS because they are smaller targets. We hypothesise that overstorey trees had lower OSS severity than tall regeneration and saplings because overstorey trees tend to have thicker bark, which may inhibit OSS's ability to feed on larger aspen stems. For example, Just et al. (2020) hypothesised that thick bark of older red maples (*Acer rubrum* L.) may inhibit feeding of gloomy scale (*Melanaspis tenebricosa* Comstock). On the other hand, overstorey trees may have had lower OSS severity than intermediate-sized stems because the OSS severity rating system does not assess OSS above 6 m in height. The mechanisms underlying OSS's outsized impacts on intermediate-sized, recruiting stems merit further research, but the potential impacts of this finding are clear. Recruiting stems are critical indicators of aspen community resilience (Rogers and Mittanck 2014; Rogers 2017), and increased mortality of these stems from OSS poses a major threat to sustainability of aspen ecosystems (Crouch et al. 2023, 2024b).

Factors influencing OSS infestation rate

Climate was the most important factor driving OSS invasions of aspen ecosystems in Arizona. According to random forests, seven of the top 10 and 15 of the top 25 factors influencing OSS infestation rate were climate variables. Moreover, SEM indicated that climate variables, namely autumn evaporation and maximum winter temperature, had the strongest direct effect on infestation rate. Generally, warmer and drier conditions were associated with increased OSS. For example, greater OSS infestation rate was associated with fewer degree-days below 0 °C and more degree-days between 10 °C and 40 °C, with warmer temperatures in winter, spring and summer, with less winter precipitation and annual snowfall and with greater spring climate moisture deficit (CMD) and winter evaporation (Table 6). The strong relationship between climate and OSS infestation rate is not surprising because of the well-established influence of climate, particularly temperature, humidity and precipitation, on development of armoured scale insects (Beardsley and Gonzalez 1975). For example, degree-day models are commonly used to predict armoured scale development (Miller and Davidson 2005a), and warmer temperatures tend to increase development rate, survival and fecundity of scale insects (Frank 2020 and references therein).

Alternatively, the relationship between climate and OSS might be mediated through host stress (Crouch et al. 2021), as indicated by the water and drought stress variables we found to be important, such as precipitation, evaporation, CMI and CMD. The plant stress hypothesis would suggest that drought not only weakens aspen defences but also increases nutritional quality, leading to increased OSS fitness and abundance (White 1984; Dale and Frank 2017; Frank 2020). In support of this hypothesis, the optimal host for sap-feeding herbivores is one that has experienced long-term, intermediate drought stress punctuated by temporary releases from that stress (Kolb et al. 2016). Our findings seem to align with this hypothesis because, although arid conditions in general were associated with more OSS, we

found that wetter conditions in autumn (e.g. reduced evaporation and increased precipitation) were associated with an increased OSS infestation rate. Therefore, we hypothesise that OSS thrives when conditions are consistently arid with temporary releases from drought in autumn. Further research is needed to assess this hypothesis and elucidate the mechanisms underlying climate's influence on OSS.

Although previous research has indicated that elevation is an important limiting factor for OSS (Crouch et al. 2021, 2024b), our study revealed that climate drives this relationship. Based on SEM, elevation did not have a significant direct effect on plot-level OSS infestation rate when climate was accounted for in the model. Instead, climate variables had significant direct effects on OSS infestation rate and were significantly correlated with elevation. A clear elevation threshold was observed in our study, in which no OSS was found above 2,545 m (Fig. 6a). However, thresholds were also observed for climate variables (Fig. 6b–f), indicating that there are cooler, wetter climatic conditions which are currently unsuitable for OSS and these conditions are closely correlated with elevation. We suspect that these thresholds, particularly the elevation threshold, will change as the climate continues to warm (Seager et al. 2007); however, repeated measurements of study plots are required to confirm this hypothesis.

Given OSS's hypothesised role as a sleeper species and the strong influence of climate on OSS infestation rate, our study suggests that climate change caused OSS population sizes to rapidly increase and to transition from an innocuous pest to a high-impact invasive species. We have shown that OSS is associated with more arid conditions. Therefore, we hypothesise that prolonged, record drought and warmer temperatures over the past 10–20 years (Williams et al. 2022) caused OSS populations in Arizona to awaken. Climate is generally considered the most common cause of sleeper species awakenings (Bradley et al. 2018; Frank and Just 2020), although other possible explanations exist, such a new mutualism, evolution, loss of a predator or parasite or introduction of a new genetic strain (Lockwood et al. 2005; Borden and Flory 2021; Spear et al. 2021). A new mutualism is unlikely because there are no documented mutualists associated with OSS (Griswold 1925; Miller and Davidson 2005b), whereas the other explanations are conceivable and represent fruitful areas for future research. For example, armoured scale populations are susceptible to suppression by natural enemies (Edmunds 1973; Raupp et al. 2010; Frank 2020), so release from one or more natural enemies could lead to OSS population growth. Interestingly, climate change can cause such a release by creating asynchrony between phenology of scale insects and their natural enemies (Frank 2020). Research on OSS genetics is needed to determine whether evolution or introduction of a new genetic strain of OSS may have influenced awakening of OSS populations. Due to the strong relationship between climate and OSS, we have serious concerns that OSS populations in other areas will continue awakening with continued climate warming (Seager et al. 2007), as has recently been observed in Utah, Nevada and Idaho (Williams 2021; Grady et al. 2022).

In addition to climate, fire had a strong influence on OSS. Fire strata was the third most important direct influence on OSS infestation rate based on SEM. Less recent fire resulted in significantly more OSS, suggesting that fire can be an important strategy for managing OSS. Of the 31 study plots that experienced fire in the two years prior to sampling, only two plots were infested with OSS, and the infestation rate in these two plots was low, with only 1.7% and 7.3% of aspen stems infested. In contrast, 40 of the 139 plots that had not experienced fire in the preceding 20 years were infested with OSS. Fire may be an important limiting

factor for OSS because it kills OSS both directly and indirectly, by killing hosts upon which OSS is dependent (Crouch et al. 2021). Aspen has thin bark, making it highly susceptible to fire mortality, so even low severity fire can kill overstorey aspen (Jones and DeByle 1985a; Stoddard et al. 2018) and, in turn, the OSS feeding on aspen. Alternatively, lack of OSS in areas of recent fire might simply be due to timing. It might take two or more years after a fire for stands of recruiting stems, which we know are susceptible to OSS infestation, to develop.

We also found that ungulate management strategies, which primarily consisted of fenced exclosures, resulted in significantly more OSS (Fig. 5). Although exclosures are highly effective at promoting aspen recruitment (Crouch et al. 2024b), this study supports previous evidence that OSS is more prevalent in areas of ungulate management (Crouch et al. 2021, 2024b). We hypothesise that ungulate exclosures promote OSS by directly increasing aspen density and, potentially, by indirectly reducing host vigour. Dense aspen stands provide more available host material and feeding sites for OSS and might facilitate OSS spread. High densities might also reduce host vigour through increased inter-tree competition (Ashton and Kelty 2018), thereby making individual aspen stems more susceptible to OSS. In support of this hypothesis, we found that aspen inside areas of ungulate management had greater height-to-diameter ratios compared to aspen outside these areas. Dense stands produce stems with greater height-to-diameter ratios (i.e. increased slenderness) (Wang et al. 1998; Frey et al. 2004), likely because trees prioritise height growth over diameter growth to compete with their neighbours for sunlight. We expect that this is especially true for shade-intolerant species such as aspen (Perala 1990). Frey et al. (2004) hypothesised that slender aspen in dense stands are more susceptible to stressors because they have more difficulty with hydraulic conductivity and may be more vulnerable to water stress and reduced photosynthesis as the stand naturally self-thins. In our study, aspen with greater height-to-diameter ratios (i.e. taller, thinner stems) were significantly associated with increased OSS presence and severity (Table 7), indicating that slender stems with reduced vigour may be more susceptible to OSS. Research is needed to determine which of these mechanisms explains why more OSS is found inside ungulate exclosures. For example, Lindroth et al. (2023) found reduced levels of phenolic glycosides, which is aspen's primary chemical defence against ungulate and insect herbivory, in unbrowsed aspen found inside exclosures, so perhaps exclosures alter aspen growth-defence trade-offs in a way that increases susceptibility to OSS.

Management implications

OSS is already widespread across several States in the Interior West, including Arizona, so management tactics intended to eradicate this pest are unlikely to succeed. Eradication is further complicated by OSS's ability to infest an array of different host species and by the fact that small populations are exceedingly difficult to detect due to OSS's small size and cryptic colouring (Crouch et al. 2021). Instead of eradication, management resources may be better spent suppressing OSS population sizes, mitigating damage to native ecosystems through integrated pest management and developing slow-the-spread tactics (Sharov et al. 2002). Robust monitoring to assess an invasive species' extent and impacts is a critical first step for successful management. Continued monitoring in areas of Arizona where we did not find OSS (e.g. southern Arizona, North Kaibab and White Mountains) is necessary, as

is continued monitoring of areas where OSS already occurs to document potential changes to the species' range (i.e. will OSS migrate to higher elevations in the future?). We also recommend monitoring of OSS in nursery stock of all host species, in urban areas that contain aspen, in riparian areas that connect urban and wildland aspen populations and in wildland aspen ecosystems, all of which may harbour sleeper populations of OSS (Frank and Just 2020; Crouch et al. 2021). Our findings can help managers identify stands at risk and prioritise which aspen ecosystems to monitor. For example, OSS is most likely to occur on warmer, drier sites, in areas that have not recently experienced fire and in stands that have dense recruitment, such as those inside fenced ungulate exclosures. Specifically, the climatic and elevation thresholds we identified (Fig. 6) can guide monitoring efforts. Although elevation is the easiest metric for managers to consider when searching for OSS, the 2,545 m threshold is liable to change across aspen's expansive range. Instead, the climate thresholds are likely to be more consistent outside our study area, though we anticipate such climate-based thresholds will shift as warming continues. These climate data can be easily obtained via ClimateNA (https://climatena.ca/).

Our findings can also be used to guide management that seeks to suppress OSS populations and mitigate damage to aspen ecosystems. Our study indicates that three strategies might help to suppress OSS populations: (1) increasing application of fire at the landscape scale, (2) reducing reliance on ungulate exclosures and (3) decreasing aspen stand density. Fire has a negative influence on OSS, and although frequency and size of wildfires will likely continue to increase as climate warming continues (Seager et al. 2007; Singleton et al. 2019), managers may consider implementing prescribed fire to suppress OSS infestations in individual aspen stands. Frequent, low-severity fire may be detrimental to aspen stands, especially in the presence of chronic ungulate browse (Crouch et al. 2023), so fire should be implemented with caution and infrequency when managing for OSS. Advantages of fire as an OSS management strategy include low cost of implementation, the ability to kill OSS on multiple host species including understorey plants and the fact that fire is a natural component of aspen ecosystems (Crouch et al. 2021). Prescribed fire may also be an effective strategy for slowing the spread of OSS from newly-discovered invasion sites.

Another strategy that managers may consider is reducing use of fenced ungulate exclosures. Reducing reliance on exclosures should help reduce OSS population sizes, although this will require finding other ways to overcome chronic ungulate browse that threatens aspen ecosystem resilience, adaptive capacity and sustainability (Rogers 2017; Crouch et al. 2023, 2024b). Dense stands with abundant recruiting stems are the structure that seems most favourable for OSS population growth. However, it remains unclear whether the relationship between aspen stand density and OSS is due to increased host availability, facilitated spread amongst densely growing stems and/or decreased host vigour in more slender stems. Further research is needed to determine how stand density and growth versus defence trade-offs influence aspen susceptibility to OSS at different spatial scales.

A third strategy managers may consider for suppressing OSS populations is reducing aspen stand densities via thinning. Thinning might also promote aspen resistance to drought, as reduced growth rates which occur in dense stands are associated with increased mortality during drought (Kane and Kolb 2014; Ireland et al. 2014, 2020; Crouch et al. 2023). However, thinning must be implemented with care in aspen stands because aspen is sensitive to mechanical damage from logging equipment

and susceptible to subsequent infection by canker-causing fungi and decay diseases (Walters et al. 1982; Jones and Shepperd 1985). Another potential drawback to thinning is that sudden, direct exposure of aspen stems to sunlight may cause sunscald (Krasnow et al. 2012), creating potential infection courts for pathogens. Before we can wholeheartedly recommend use of prescribed fire or silvicultural strategies like thinning to manage OSS, long-term experimental research is needed to assess the efficacy of these strategies for suppressing OSS populations and mitigating damage to aspen ecosystems. Research is also needed to assess the efficacy of other forms of management, such as biological control and application of systemic insecticides, that have no direct negative effect on aspen and will be critical components of an integrated pest management programme for OSS. Our future work seeks to address these needs by assessing OSS and aspen responses to various suppression strategies.

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Additional information

Conflict of interest

Ethical statement

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Author contributions

Conceptualization: RWH, KMW, CDC, AMG, MMM, NPW. Data curation: CDC, KMW. Formal analysis: CDC. Funding acquisition: MMM, CDC, AMG, KMW, NPW. Investigation: CDC, AMG, KMW, NPW. Methodology: MMM, CDC, AMG, NPW, RWH, KMW. Project

administration: AMG, CDC, NPW, KMW. Resources: NPW, AMG, KMW. Supervision: KMW. Validation: CDC. Visualization: CDC, KMW. Writing – original draft: CDC. Writing – review and editing: CDC, MMM, KMW, NPW, RWH, AMG.

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Data availability

All of the data that support the findings of this study, including R code, are available via the Environmental Data Initiative (https://doi.org/10.6073/pasta/bd7be772e435ed0ba5585aae5a96f3e7).

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Supplementary material 1

Means and standard errors for the 92 continuous variables considered as potential influencing factors of plot-level OSS abundance, summarized across each of the seven major areas in our study

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